STUDIES IN THE METABOLISM OF APPLES.

V. THE RESPIRATORY METABOLISM OF DELICIOUS APPLES OF COMMERCIAL MATURITY AFTER VARIOUS PERIODS OF COOL STORAGE.

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(Six Text-figures.)

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Introduction.

Previous papers in this series (Hackney, 1943*a*, 1943*b*, 1944) are concerned with the respiratory metabolism of Granny Smith apples after various periods of cool storage. It was shown that in Granny Smith apples which had not been in store for more than three or four months internal oxygen supply plays a very important part in the limitation of the respiratory activity. Evidence already collected indicates that the changes in internal oxygen concentration are affected by such factors as number of lenticels and their degree of openness, and the amount of oil developed on the skin. The present investigations were undertaken on Delicious apples in order to compare their metabolism with that of Granny Smith apples. Delicious apples differ from Granny Smith apples in that their skins generally do not develop very much oil. There are also important anatomical differences between the two varieties.

The present paper embodies the results of investigations on the post-storage metabolism of Delicious apples of commercial maturity in the 1942 and 1943 seasons. The rôle of oxygen in the metabolism of Delicious apples is very different from that taken in the metabolism of Granny Smith apples, and it will be shown that the difference is probably related to the difference between the fruits in their permeability to gaseous diffusion.

Materials and Methods.

The fruits for the 1942 experiments were part of the normal commercial picking from Batlow, N.S.W. They were picked on 25.iii.42 and placed in cool store at Batlow until 20.v.42. They were then despatched to Sydney, where they were placed again in cool store (0°C.) one day later (21.v.42). At intervals of two weeks from 26.v.42, samples of five fruits each were withdrawn from store and taken to a room at a constant temperature of 21°C. For each apple in the sample, the rate of output of carbon dioxide and the composition of the internal atmosphere were measured at regular intervals, the measurements being continued in most samples until the fruits showed signs of advanced senescence.

Rates of output of carbon dioxide were measured by the Pettenkofer method and expressed in milligrams of carbon dioxide per 10 Kilograms fresh weight per hour. Concentrations of oxygen and carbon dioxide in the internal atmosphere were measured, using the method described in the first paper of this series (Trout *et al.*, 1942).

The fruits for the 1943 experiments were picked at Orange, N.S.W., on 15.iv.43. They were stored in Sydney at 0°C. on 19.iv.43. Samples of five fruits each were withdrawn from store at fortnightly intervals. The first sample was taken on 19.iv.43 and had not been in cool store. The subsequent behaviour of all the samples was observed at 18·3°C.,

no room at 21°C. being available. Measurements were made of the rates of uptake of oxygen and output of carbon dioxide and of the concentrations of these gases in the internal atmosphere.

The Pettenkofer method was not used in 1943. The rates of uptake of oxygen and output of carbon dioxide were measured by the method described by Sykes (1944).

Shortly after investigations were begun in 1942 on Delicious apples, it was discovered that about 75% of the fruits had open calices—a feature almost completely absent from Granny Smith apples. The condition of the calyx was ascertained as follows: The fruit, fitted with a gas-sampling pipette,* was immersed in water, the end of the pipette being held in air. The pipette was then opened temporarily while air was blown gently through the fruit. Bubbles appeared through the calyx if it was open. While carrying out this test it was discovered that the lenticels of Delicious apples are very frequently open. Those of Granny Smith apples are generally closed. As the investigations were planned in order to compare the metabolism of two varieties (Granny Smith and Delicious) having different types of skin, it was thought best in 1942 to seal the open calices with wax and so limit gaseous diffusion to the skin.

During 1943, the calices were sealed in samples 1 and 2, but in later samples they were left untreated. The degree of closure was ascertained by the air bubble technique used in 1942. If no bubbles appeared when air was blown into it, the fruit was classified as 'closed'; if few bubbles appeared, it was classified as 'half-open'; if a rapid stream of bubbles appeared, it was classified as 'open'. When being analysed, the results were grouped according to these three categories.

Experimental Results. A—1942 Season.

Table 1 shows date of removal from store, date of insertion of gas-sampling pipette, date of first observation, date of final observation and number of weeks in cool store for each of the twelve samples taken during 1942.

Table 1.

Delicious Apples of the 1942 Season; Picked 25.iii.42; Cool-stored at 0° C. on 27.iii.42.

Sample Number.	Date of Removal from Store.	Date of Insertion of Sampling Pipette.	Date of First Observation.	Date of Final Observation.	Number of Weeks in Cool Store.
1	26.v	26.v	27–28.v	27–29.vi	9
2	8.vi	9.vi	10–11.vi	23–24.vii	11
3	22.vi	23.vi	24–25.vi	17–18.viii	13
4	7.vii	8.vii	9–10.vii	11–12.viii	15
5	20.vii	21.vii	22–23.vii	22–24.viii	17
6	3.viii	4.viii	5– 6.viii	1– 2.ix	19
7	19.viii	19.viii	20–21.viii	15–16.ix	21
8	2.ix	3.ix	4– 5.ix	4– 5.xi	23
9	22.ix	23.ix	24–25.ix	3– 4.xi	26
10	12.x	12.x	13–14.x	5– 6.xi	29
11	26.x	26.x	27–28.x	12–13.xi	31
12	10.xi	10.xi	11–12.xi	30.xi–1.xii	33

Respiration Rates.—Figure 1 shows mean rate of output of carbon dioxide for each of the twelve samples. Owing to technical difficulties readings were not obtained for sample 4 until fifteen days after removal from store. With the exception of those of sample 5, all the fruits of all the samples had very high rates of output of carbon dioxide during the first few days after removal from store. Considerable variability was observed between the respiration rates of individuals within each sample; therefore the standard deviation of each sample was very large. However, the shape of the respiration-time curve was the same for each fruit. In general, the mean respiration rate fell irregularly throughout the period of observation. The peak observed in sample 1 was

* The pipette had been inserted in order to sample the internal atmosphere.

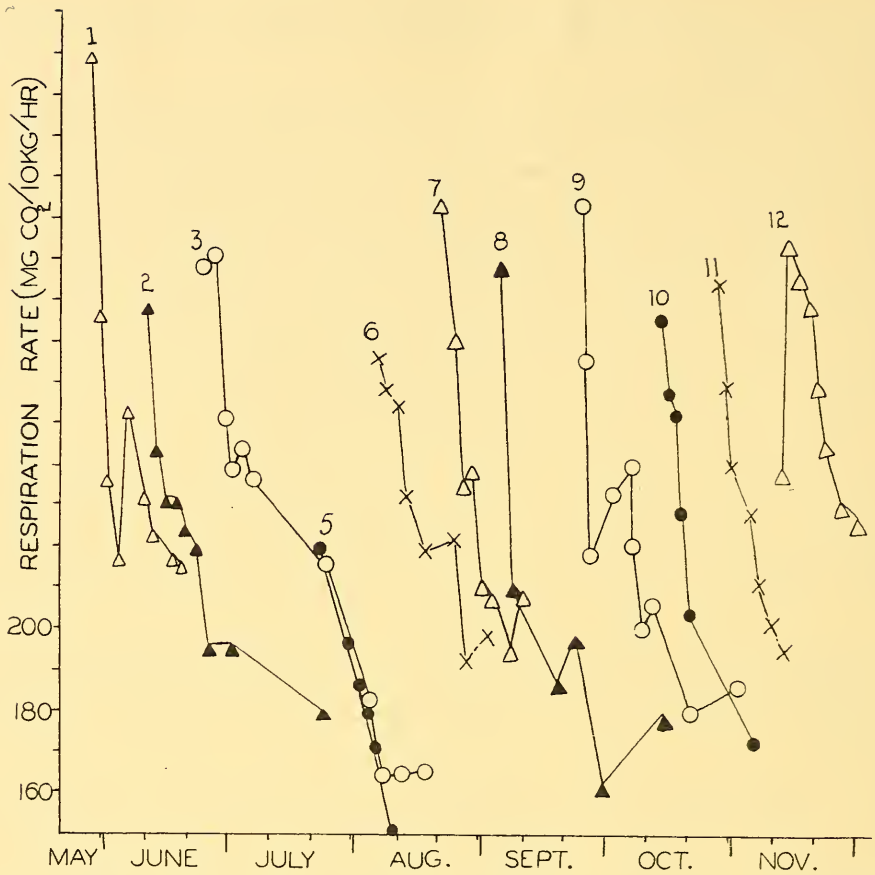

Fig. 1.—Mean rates of output of carbon dioxide for eleven of the twelve samples of Delicious apples taken during 1942. Values for sample 4 have been omitted to avoid confusion of the figure.

significant;* it might have been due to the fact that the fruits were sealed only a few hours before the highest respiration rate was observed. The open calices were not noticed until 4.vi.42. The equilibrium between the gas concentrations inside and outside the fruits might not have been regained when the reading was taken. With the exception of sample 5, where the variability was unusually low, no differences were observed between the mean initial respiration rates of the various samples.

Composition of the Internal Atmosphere.—Figure 2 shows the mean concentrations of oxygen and carbon dioxide in the internal atmosphere for each sample taken during 1942. The calices of fruits in sample 1 were sealed after observations had been made on 5.vi.42. In the early samples (1 to 8 inclusive), the mean internal oxygen concentration fell throughout the period of observation, the lowest values being attained by samples 4, 5 and 6. Very little change was observed in the corresponding concentrations of carbon dioxide. The initial internal oxygen concentration showed a tendency to decrease from sample to sample until sample 5 was reached. After this no definite trend was observed. Sample 5 had the lowest internal oxygen concentration. In the late samples (9 to 12 inclusive), the internal oxygen concentration was comparatively high

* The peak observed in sample 1 was significant because it occurred in every individual fruit of the sample. The peak observed in the mean respiration rate of sample 12 was not significant since the values for some fruits of this sample remained low while those for the other fruits rose, i.e., the standard deviation was high at this point of the curve.

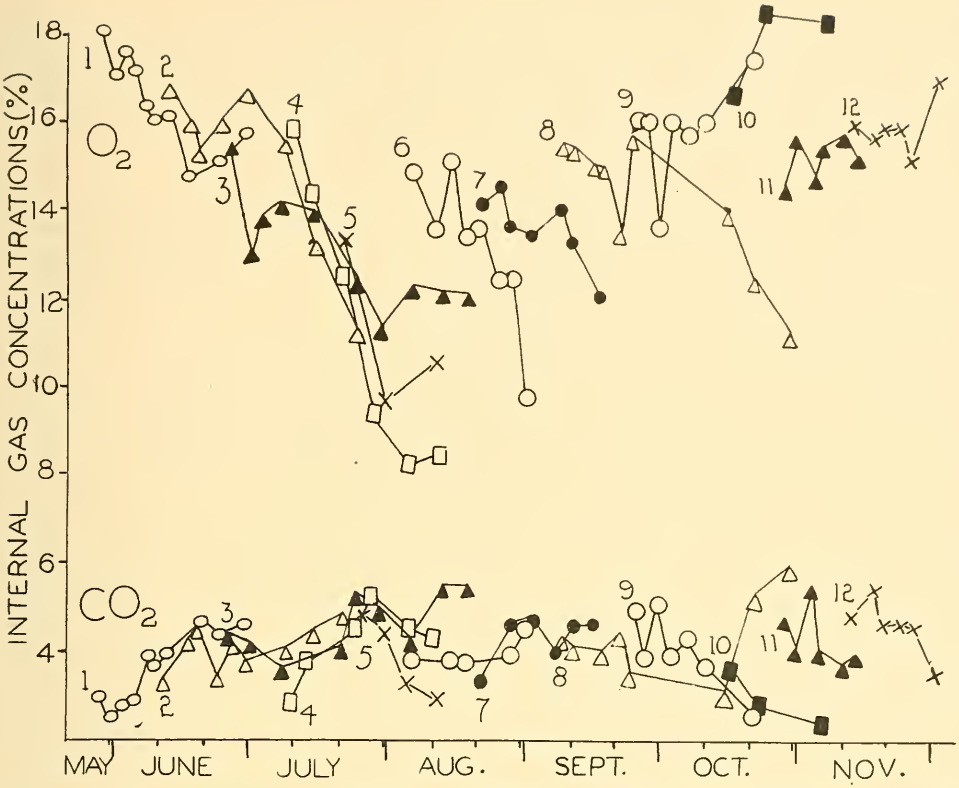

Fig. 2.—Mean concentrations of oxygen and carbon dioxide in the internal atmospheres of the twelve samples of Delicious apples taken during 1942.

initially and either remained constant or rose slightly. The corresponding concentrations of carbon dioxide generally fell throughout the period of observation.

Resistance of the Skin to Gaseous Diffusion.—The resistances of the skin to the diffusion of oxygen (r_o) and carbon dioxide (r_c) were calculated by the method outlined in the first paper of this series (Trout *et al.*, 1942). The mean values for the various samples are shown in Figure 3. In samples 1 to 7, r_o increased throughout the period of

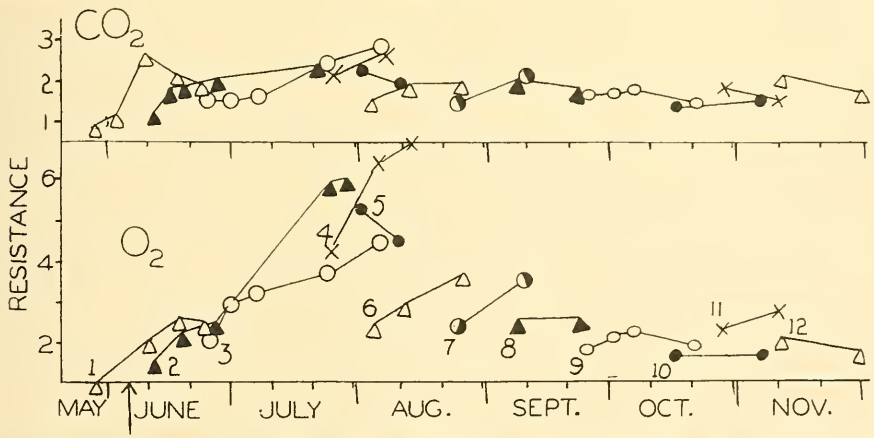

Fig. 3.—Mean resistances to the diffusion of oxygen and carbon dioxide for each of the twelve samples of Delicious apples taken during 1942. The arrow indicates date of closure of the open calices.

observation, the increase being most marked in samples 1 to 4 (over 4 units in 40 days in sample 2). In samples 8 to 11 inclusive, r_o remained constant throughout the period of observation. The initial values for r_o increased from sample to sample until sample 5 was reached. This sample had the highest initial value for r_o. The initial values for r_o were comparatively low in samples 6 to 12 inclusive.

The value for r_c showed very little change throughout the year.

Experimental Results. B—1943 Season.

Table 2 shows date of removal from store and insertion of gas-sampling pipette (same day), date of first observation, date of final observation and number of weeks in cool store for each of the sixteen samples taken.

Table 2.

Delicious Apples Picked 15.iv.43; Cool-stored (0° C.) on 19.iv.43.

Sample Number.	Date of Removal from Store.	Date of First Observation.	Date of Final Observation.	Number of Weeks in Cool Store.
1	19.iv	20.iv	18.v	0
2	4.v	5.v	11.vi	2
3	18.v	19.v	13.vii	4
4	1.vi	2.vi	4.viii	6
5	15.vi	16.vi	30.vii	8
6	6.vii	7.vii	17.viii	11
7	20.vii	21.vii	10.ix	13
8	2.viii	3.viii	2.ix	15
9	18.viii	20.viii	8.ix	17
10	31.viii	1.ix	21.x	19
11	15.ix	16.ix	21.x	21
12	5.x	6.x	18.x	24
13	19.x	20.x	11.xi	26
14	2.xi	3.xi	23.xi	28
15	16.xi	17.xi	1.xii	30
16	7.xii	8.xii	13.xii	33

Respiration Rates.—For the preliminary analysis of the data, the fruits were grouped in three anatomical classes—those with closed calices, those with moderately open calices and those with very open calices. The rates of uptake of oxygen and output of carbon dioxide were then considered separately for each apple in each group. Great variability was observed between individual fruits of the same type in the same sample, and no significant differences were observed between individual fruits of the same type in different samples or between those of different types in the same sample. In view of these facts, mean values have been calculated for all the fruits of each sample, and these are given in Figure 4. The values for some of the samples have been omitted in order to make the figure clear. Nothing essential is lost by doing this, as the samples omitted behaved similarly to those included. The rates of uptake of oxygen and output of carbon dioxide usually fell irregularly throughout the period of observation, but slight significant peaks (see footnote, p. 110) were observed in the rates of output of carbon dioxide in samples 4, 5, 6 and 7 soon after removal from store. Similar peaks were also observed in occasional individual fruits of other samples. Their occurrence was not limited to apples of any anatomical class. The mean initial rates of uptake of oxygen and output of carbon dioxide showed no significant trends from sample to sample. The rates of fall in respiratory activity were approximately the same for all the samples.

The Respiratory Quotient.—Mean values for respiratory quotients of the sixteen samples are shown in Figure 4. Only the first few values obtained are included in the figure as no change occurred subsequently. As in Granny Smith apples of the 1943 season (Hackney, 1944), some of the samples had respiratory quotients of less than 1 on the day after removal from store. The respiratory quotient had generally risen to approximately 1 when the second observation was made. It remained at this level during the remainder of the period of observation. There is no reason to suppose that the first observation

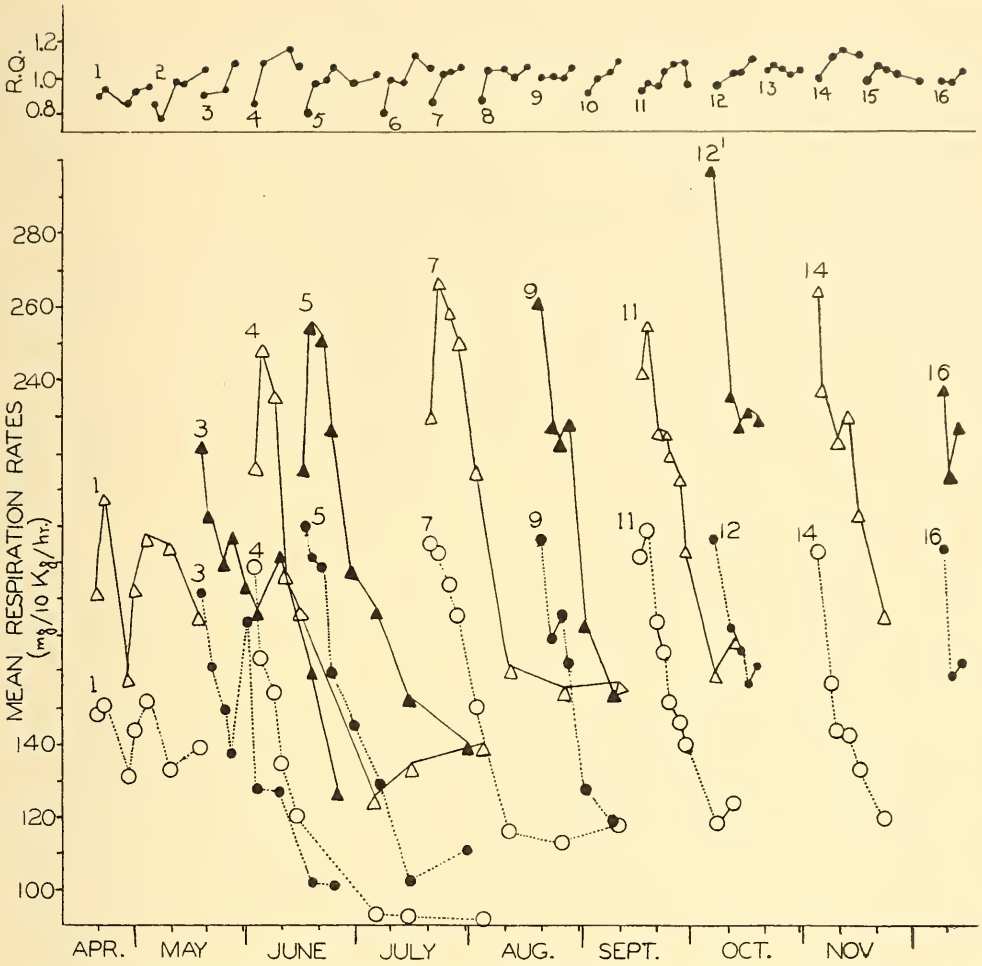

Fig. 4.—Mean values for respiratory quotient (R.Q.) for all the samples, and mean respiration rates for ten of the sixteen samples of Delicious apples taken during 1943. The samples omitted behaved similarly to those included. Values for rate of uptake of oxygen are joined by dotted lines; those for rate of output of carbon dioxide are joined by entire lines.

after removal from store was more subject to technical inaccuracies than subsequent observations (see Hackney, 1944).

Composition of the Internal Atmosphere.—For the comparison of the compositions of the internal atmospheres the fruits were grouped in three anatomical classes, according to the structure of the calyx. Figure 5*a* shows the internal concentrations of oxygen in the fruits which had open calices (means for each sample). The mean internal concentrations of carbon dioxide in these fruits were low (about 3% or less). During the period of observation at 18·3°C. very little change occurred in the composition of the internal atmospheres of these fruits. Further, the composition of the internal atmosphere on removal from store was the same at the end of the year as at the beginning. The internal atmospheres of fruits with moderately open calices were similar in composition to those of fruits with very open calices. When, however, the mean internal oxygen concentrations for all the fruits with closed calices were compared with those for fruits with open calices, the differences were statistically significant ($P < 0{\cdot}001$). When the behaviour of individual fruits was considered, it was found that fruits with closed calices varied in the composition of their internal atmospheres. In

some fruits of this type the internal atmospheres were not significantly different from those of fruits with open calices. In other fruits the internal oxygen concentration was either initially low or fell during the period of observation until it reached a level considerably lower than the lowest level observed in any fruit with an open calyx.

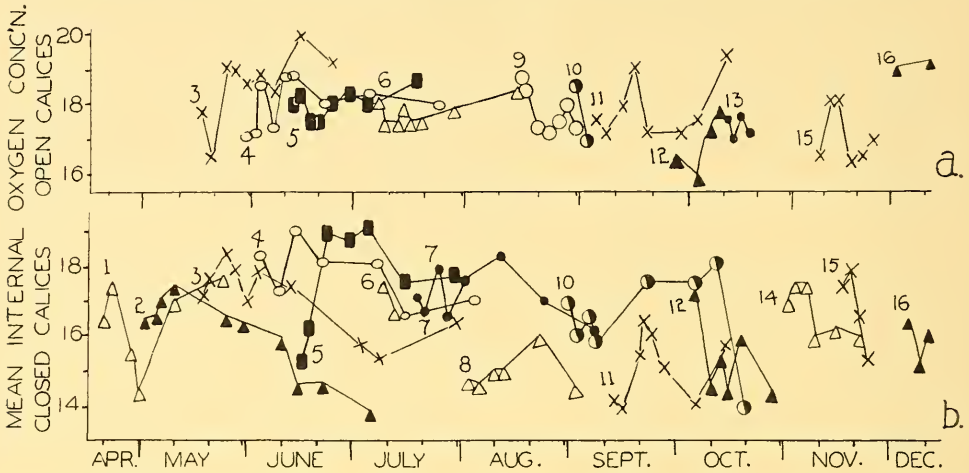

Fig. 5.—Mean internal oxygen concentrations for (*a*) fruits with open calices, (*b*) fruits with closed calices. The fruits were those of the 1943 season, and the numbers on the curves indicate the samples from which they were taken.

Figure 5*b* shows the mean internal oxygen concentrations of the fruits with closed calices. No significant difference was observed between the mean concentrations of carbon dioxide in the internal atmospheres of fruits of the three anatomical classes.

Resistance to Gaseous Diffusion.—In fruits with open calices the values for resistance to the diffusion of oxygen were low, and very little change was observed throughout the period of observation. Figure 6 shows the corresponding values for fruits with closed calices. In some of these fruits no appreciable change was observed during the period of observation. In others the resistance to the diffusion of oxygen rose considerably after removal from store. A striking example of variability is pre-

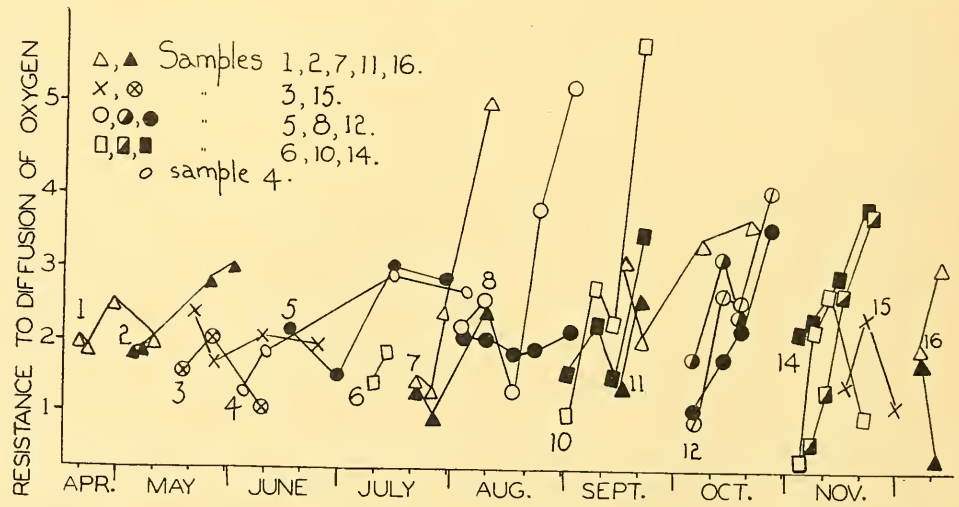

Fig. 6.—Values for resistance to diffusion of oxygen for individual fruits which had closed calices in 1943. The numbers on the curves indicate the samples from which these fruits were taken; each fruit is represented by a different symbol.

sented by the two 'closed' fruits of sample 8. The oxygen resistance of one of these remained constant throughout, while that of the other rose steeply.

In most fruits the resistance to the diffusion of carbon dioxide did not change appreciably, but it increased considerably in a few fruits which had closed calices (e.g., in sample 12, original values for three fruits were 1·1, 1·2 and 1·2 units, respectively, and final values were 4·1, 3·6 and 2·4, respectively).

Discussion of Results.

A. Comparison of results for 1942 and 1943.

Respiration Rates.—In many respects the respiration records of the 1943 fruits are similar to those of the 1942 fruits. In both seasons the mean rates of output of carbon dioxide for any particular sample on successive occasions generally fell throughout the period of observation. Some of the 1943 samples showed slight respiratory peaks, the occurrence of which could not be correlated with any particular characteristic of the fruit. No definite trends were observed among the initial respiration rates of successive samples in either season. The general level of the respiration rate was higher in 1942 than in 1943, probably because the fruits were held at a higher temperature (21°C.) during 1942.

In 1942, alcohol analyses were carried out on fruits which had been in store for various periods. The analyses were carried out about six days after removal from store and it was found that the fruits contained between 0·02% and 0·05% of alcohol. In 1943, it was found that the respiratory quotient was approximately 1 when the fruit had been out of store for three days or longer. If any anaerobic respiration was going on, the amount of carbon dioxide produced in this way was apparently too small to change the respiratory quotient noticeably.

Composition of the Internal Atmosphere.—Changes in the composition of the internal atmosphere with time were much more marked in 1942 than in 1943. Two factors probably contributed to this difference. In 1942 all the fruits had closed calices, those which were naturally open being sealed with wax, whereas in 1943 this was not so. Thus the population was more homogeneous in 1942 than in 1943. In addition, the experimental temperature was nearly three Centigrade degrees higher in 1942. As a general rule, any changes in the internal atmosphere of an apple occur more quickly and are more pronounced at higher than at lower temperatures. In 1942 the samples fell into two classes: those which had been in store for less than six months and those which had been in store longer. In the former class the internal oxygen concentrations of all the fruits fell throughout the period of observation, the lowest mean value observed being 8·3%. In the latter class the internal oxygen concentrations remained approximately constant. In 1943 the mean behaviour of all the samples was the same. In fruits which had open calices the internal oxygen concentration was high (about 18–19%) and the concentration of carbon dioxide was low (about 2–3%). In fruits with closed calices the extent of the changes in the composition of the internal atmosphere varied considerably. In some fruits the internal concentration of oxygen was high (about 18%) and did not change during the period of observation. In others the internal oxygen concentration was high initially and fell slowly at 18·3°C. In the remaining fruits the internal oxygen concentration was low initially (lowest observed being 10·8%) and either rose or remained constant subsequently. Thus in 1943 the group which most closely compared with the fruits of 1942 (i.e., the closed calyx type) showed considerably more variability within samples than did the 1942 fruits. It was observed that whereas in 1942 the earlier samples (1 to 8 inclusive) became very greasy after removal from store, the development of grease was not so great in the 1943 samples. Experience indicates that this difference may be due to the higher experimental temperature in 1942. In 1943 the lenticels were more open in some fruits than in others. Those with very open lenticels always had high internal oxygen concentrations. In 1942 the lenticels were open but did not appear to be as widely open as those of some of the 1943 fruits.

Resistance to Gaseous Diffusion.—Changes in the resistance to gaseous diffusion were more marked in 1942 than in 1943. In the early samples of the 1942 season, which were greasy, the resistance to the diffusion of oxygen (r_o) increased throughout the period of observation. The initial values for r_c were low (about 2 units) and subsequent changes were small or negligible. Later samples were not noticeably greasy. In addition to the normal samples 8 and 9 (1942), in which the calices were waxed, two extra samples were taken from store on the same days as samples 8 and 9, respectively. The calices of these fruits were left open. There was no difference between these samples and samples 8 and 9 in respiration rate, composition of the internal atmosphere, or changes in r_o and r_c. In the early 1942 samples, where the value for r_o was high, any leaks which developed in the wax seals caused the internal oxygen concentration to rise noticeably. It would seem that in 1942 the resistance of the skin of the late samples was so low that the closure of the calyx made little difference.

In the 1943 samples, the resistance was always low (1 to 3 units) in fruits with open calices. In fruits with closed calices the values for r_o and r_c were sometimes so low as to be equal to those for fruits with open calices. In the majority of fruits with closed calices the value for r_o rose during the period of observation. As in the late samples in 1942, resistance of the skin was occasionally so low that the closure of the calyx made no noticeable difference.

B. GENERAL DISCUSSION.

Respiration Rates.—With the exception of the slight initial peaks observed in some of the early 1943 samples, there was no suggestion of any rise in respiration rate accompanying the onset of ripening. In 1942 the early samples were fairly ripe when removed from store, but in 1943 they were still flavourless. Fisher (1943) found that in Delicious apples at 60°F. (15·5°C.) respiration rate reached its peak about five days after picking. As the fruits considered in this paper were not put into store until several days after picking, possibly the respiratory peak was missed. Whether the slight peaks observed in the rates of output of carbon dioxide in samples 4, 5, 6 and 7 in 1943 were analogous to the climacteric peaks of Blackman and Parija (1928) and other workers or not, no peaks were observed in the corresponding curves for rate of uptake of oxygen; it is probable that most of the fruits of the 1943 season passed from the unripe to the fully senescent stage without showing any evidence of an increase in respiratory activity, measured either as uptake of oxygen or output of carbon dioxide. This probability, together with the fact that the Granny Smith apple has frequently been observed to ripen without any associated rise in the rate of respiration (Hackney, 1943), suggest that the 'climacteric peak' is not nearly as common an accompaniment of the onset of ripening as was once supposed.

In several of the 1943 samples the respiratory quotient, though approximately 1 for the greater part of the storage life, was significantly less than 1 on the day after removal from store. The reason for this is not known. The possible reasons for the similar phenomenon in Granny Smith apples have been discussed in the previous paper (Hackney, 1944).

Resistance of the Skin to Gaseous Diffusion, and Composition of the Internal Atmosphere.—In spite of the variations in resistance to gaseous diffusion, certain principles of behaviour appear to have been common to fruits of 1942 and 1943. The extent to which changes in the resistance of the skin affect the composition of the internal atmosphere depends on the extent of closure of the calyx. If the calyx is open no changes occurring naturally in the resistance of the skin impose sufficient restriction on the rate of gaseous diffusion to cause any noticeable change in the composition of the internal atmosphere. If the calyx is completely or almost completely closed, changes in the resistance of the skin tend to modify the composition of the internal atmosphere. In some of the closed fruits no noticeable changes occur in the resistance of the skin. It has been observed that these fruits frequently have very open lenticels, as measured by the pressure required to blow air through them. During 1943, the value for r_o

increased throughout the period of observation when the calyx was closed. The increase in r_o was generally accompanied by a corresponding increase in r_c.

It appears that there is no invariable correspondence between the degree of closure of the calyx and the composition of the internal atmosphere. Late in 1943, four fruits were selected in which the calices were completely closed. Observations were made on rates of uptake of oxygen and output of carbon dioxide and composition of the internal atmosphere while the calices were closed. The calices were then opened with a sterilized needle and a similar set of observations was made 24 hours later. The concentrations of oxygen and carbon dioxide in the internal atmospheres before and after the opening of the calices are shown in Table 3. Corresponding values for r_o and r_c were calculated, assuming that the respiration rates of the fruits were approximately the same before and after the opening of the calices. In the two fruits (numbers 1 and 3 in Table 3), which had the highest values for r_o and r_c (about 2·0 and 1·6 units, respectively), the opening of the calices resulted in a noticeable reduction of both values (2·0 $\longrightarrow$ 1·1 units; 1·6 $\longrightarrow$ 1·0 units). In the other two fruits, where the initial resistances were slightly lower (r_o = about 1·4 units; r_c = about 1·3 units), the differences consequent upon the opening of the calices were so slight as to be insignificant; that this might have been due to the openness of the lenticels is shown by a second experiment (see below).

TABLE 3.

Composition of the Internal Atmosphere before and after Artificial Opening of the Calices of Delicious Apples in December, 1943.

Fruit No.	Date.	Condition of Calyx.	Internal O_2 (%).	Internal CO_2 (%).
1	30.xii.43	Naturally closed.	16·9	2·9
2	,,	,, ,,	18·7	2·1
3	,,	,, ,,	16·9	3·2
4	,,	,, ,,	17·7	2·9
1	31.xii.43	Artificially opened.	18·7	2·0
2	,,	,, ,,	18·9	1·8
3	,,	,, ,,	19·2	2·1
4	,,	,, ,,	18·0	2·6

In June, 1944, six Delicious apples were selected having closed calices and very open lenticels. They were placed in a room maintained at a constant temperature (21°C.). The concentrations of oxygen and carbon dioxide in the internal atmospheres were determined on 26.vi.44. The lenticels of three of the fruits were then painted with a solution of castor oil and shellac in alcohol, one of the preparations which are known to increase resistance to the diffusion of oxygen when applied to the skin (see Hackney, 1943*b*). The concentrations of oxygen and carbon dioxide in the internal atmospheres were again measured on 13.vii.44. The internal oxygen concentrations had decreased considerably in the fruits whose lenticels had been painted; in the untreated fruits the changes were small or negligible (Table 4).

It appears that in some fruits with closed calices the resistance of the skin is so low (probably due to the openness of the lenticels) that the composition of the internal atmosphere is not noticeably different from that in fruits with open calices.

Comparison between Granny Smith and Delicious Varieties.—When the data presented in this paper are compared with those previously presented for Granny Smith apples (Hackney, 1943*a*, 1943*b*, 1944) it is obvious that there are differences in behaviour between the two varieties. Mature Delicious apples have much higher respiration rates than Granny Smith apples under the same conditions. Changes in the composition of the internal atmosphere and in the resistance of the skin to gaseous diffusion are frequently less marked in the former variety than in the latter. When the preliminary investigations were being carried out on Granny Smith apples, several tests were carried out which proved that the resistance of the flesh of the fruit is very small compared with that of the skin (Trout *et al.*, 1942). Similar tests carried out on Delicious apples

TABLE 4.

Composition of the Internal Atmosphere before and after Artificial Closure of the Lenticels of Delicious Apples in June, 1944.

Fruit No.	Date.	Condition of Lenticels.	Internal O_2 (%).	Internal CO_2 (%).
1	26.vi.44	Naturally open.	14·0	5·2
2	,,	,, ,,	14·8	5·1
3	,,	,, ,,	17·5	6·4
4	,,	,, ,,	16·6	6·0
5	,,	,, ,,	14·9	4·6
6	,,	,, ,,	16·5	4·0
1	,,	,, ,,	15·3	4·9
2	,,	,, ,,	17·9	4·2
3	,,	,, ,,	14·5	4·6
4	,,	Artificially closed.	3·9	8·6
5	,,	,, ,,	8·4	5·9
6	,,	,, ,,	11·6	7·3

in 1943 showed that in this variety also the resistance of the flesh is very low. It appears, therefore, that the differences between the resistance changes observed in the two varieties are due to differences in the skin and in the structure of the calyx. The occurrence of the open calyx in the Delicious variety and its absence from the Granny Smith have already been mentioned. In addition, the development of oil on the surface of the fruit is often more noticeable in the Granny Smith than in the Delicious apple. Probably the most important difference between the skin of the Delicious apple and that of the Granny Smith is that of the lenticel structure. It has been shown by anatomical study (N. O'Grady, 1941*) and by the air-bubble test described in this paper, that the lenticels of the Granny Smith apple are generally closed. In the Delicious apple they are generally open. It has been observed by the writer that, in the very few Granny Smith apples where the lenticels were open, there were considerably greater concentrations of oxygen in the internal atmospheres than in comparable apples where the lenticels were closed.

Data have been presented in previous papers (Hackney, 1943*a*, 1943*b*, 1944) to show that oxygen supply plays a very important part in the limitation of the respiration rate of Granny Smith apples during a certain period of their life. Strong positive correlations have been observed between internal oxygen concentration and respiration rate in Granny Smith apples which have not been in store longer than three or four months. The same apples have shown rapid acceleration of respiratory activity when held in pure oxygen. No data have been obtained regarding the behaviour of Delicious apples in pure oxygen. In the early samples of Delicious apples of the 1942 season, strong positive correlations ($P < 0{\cdot}01$) were observed between internal oxygen concentration and respiration rate, but in later samples of 1942, and in all the 1943 samples, no such correlations were observed. Respiration rate frequently fell while internal oxygen concentration remained constant. It appears that some factor other than oxygen supply limits the respiration rates of the majority of Delicious apples.

It is evident from the complexity of the results presented in this paper, and others of the same series, that investigations of the respiratory metabolism of fruits should include not only observations of the external respiration rates but detailed studies of the changes which may occur in the resistances of the fruits to gaseous diffusion. Without such studies it is impossible to decide whether changes in the respiratory intensity are due to limitation of the internal oxygen supply or to some other factor, such as substrate starvation or the deterioration of the enzyme system.

SUMMARY.

During 1942 and 1943, investigations have been carried out on Delicious apples of commercial maturity after various periods of storage at 1°C.

* Unpublished data.

Observations have been made on rates of uptake of oxygen and output of carbon dioxide, respiratory quotients and composition of the internal atmosphere. The resistances of the fruit to the diffusion of oxygen and carbon dioxide have been calculated from the data obtained.

It was found that the Delicious apple frequently has an open calyx. In an attempt to ensure uniformity, the open calices were sealed with wax in 1942; during 1943 the calices were left untreated, and the data have been analysed in three classes according to the degree of closure of the calices.

The rates of uptake of oxygen and output of carbon dioxide generally fell throughout the period of observation. In 1943, slight peaks were observed in the rates of output of carbon dioxide in some samples shortly after removal from store. No corresponding peaks were observed in the rates of uptake of oxygen. No difference was observed between the respiration rates of apples with closed calices and those with open calices.

The respiratory quotient was frequently less than 1 on the first day after removal from store, but had risen to 1 on the third day after removal from store.

The degree of closure of the calyx had a small but statistically significant effect on the mean internal oxygen concentration. In fruits with open calices the internal oxygen concentration remained high. In fruits with closed calices it sometimes fell to a relatively low level.

In fruits with open calices the resistances of the fruit to the diffusion of oxygen and carbon dioxide did not rise during the period of observation. In those where the calyx was completely or almost completely closed, the resistances to the diffusion of oxygen and carbon dioxide frequently rose.

These results are discussed in detail, and are compared with those presented in previous papers for Granny Smith apples. Attention is drawn to the great differences between the rôles of oxygen in the metabolism of the two varieties. These differences are related to differences in the permeabilities of the two varieties to gaseous diffusion.

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